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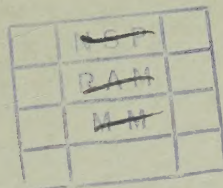
**A WITCHES' BROOM VIRUS DISEASE OF
VACCINIUM MYRTILLUS IN THE NETHERLANDS**

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A WITCHES' BROOM VIRUS DISEASE OF *VACCINIUM MYRTILLUS* IN THE NETHERLANDS¹

*Met een samenvatting: Een heksenbezemvirusziekte van
Vaccinium myrtillus in Nederland*

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INTRODUCTION

Shortly after the appearance of the author's publication on witches' broom phenomena (BOS, 1957) Dr. M. HILLE RIS LAMBERS kindly directed attention to a peculiar disease of bilberry (*Vaccinium myrtillus* L.), having many symptoms in common with witches' broom virus diseases. The disease appeared to be rather prevalent in the woods in the neighbourhood of Bennekom, a small village near Wageningen. According to Dr. HILLE RIS LAMBERS the disease occurred also at Loenermark not far from Apeldoorn.

Since the leafhopper-spread witches' broom viruses appear to be of increasing scientific and economic importance, this *Vaccinium* disease is being studied in some detail. This paper gives an account of some preliminary results of these investigations. A survey of literature concerning possibly related diseases occurring in other countries will be given in the discussion.

DESCRIPTION OF THE DISEASE

In the woods occurring on the dry sandy, soils of the „Veluwe" bilberry is a very common, wild plant species. The fruits are not harvested commercially, but are gathered on a large scale by housewives and children for the production of jam for home consumption. Diseases of this crop may therefore be of some economic importance. Since the witches' broom diseased plants are generally entirely sterile they do not produce fruits.

At present no general information exists as to the distribution of the witches' broom disease under investigation. In the neighbourhood of Bennekom the symptoms occur in large groups of plants. The way of occurrence suggests an infectious agent. The disease is more prevalent in more open parts of the wood.

Affected plants attract special attention by a very dense, broomy growth. This is due to an excessive branching, and a very erect position of the newly formed branches. The excessive ramification of the plants is associated with a striking reduction in size of branches and leaves. The leaves may have a length of 4 mm, or even smaller, instead of 15-20 mm as in normal leaves.

The final result of the disease depends on the stage of development of the plant at the time of affection (Fig. 1). In plants or plant parts affected at a late stage of growth, the branches have only an erect position instead of the plagiotropic position as in normal plants. Moreover, some additional branching may

¹ Accepted for publication 23 March, 1960.

occur. Plants affected earlier in their development remain smaller in size, are heavily branched and show a decrease in leaf size. The young plants, especially at the edge of a group of plants, remain extremely small (about 5 cm) with very small leaves. These severely diseased plants do not form subterranean suckers.

During autumn the diseased plants drop their leaves later than normal plants do. The underlying persistence of physiological activity in winter may lead to high frost damage. Early in spring the affected plants start developing new foliage and sprouts. At this time and in autumn, affected plants may be easily detected. In experiments performed in a heated glasshouse during winter healthy plants lost their leaves, whereas diseased plants went on producing branches and leaves (Fig. 2).

Generally the foliage shows a light green colour in contrast to the dark green colour of healthy plant foliage. This may be due to some vein clearing, but is especially promoted by the continual production of young new leaves and the somewhat etiolated dense growth of the brooms. Sometimes the leaves show an increased anthocyanin formation, leading to some reddening of these leaves.

Diseased plants do not flower at all and therefore no floral abnormalities could be observed. The plants are entirely sterile.

EXPERIMENTS AND RESULTS

Since the symptoms and the occurrence of the disease strongly suggest a virus to be the cause, and since no abnormalities indicate the presence of parasitic organisms, experiments have been carried out to demonstrate that a virus is involved.

During these experiments physiological disturbances, such as climatic or nutritional disorders, were ruled out as the cause of the disease. In all our experiments diseased and healthy plants were potted in the same humous, acid soil, collected in the woods. Both diseased and healthy plants were transported to the same glasshouse, where they have been treated in exactly the same way for more than one year. The affected plants continue to develop exactly the same symptoms, whereas the healthy plants remain healthy. These observations clearly exclude the presence of a non-infectious agent.

Some preliminary experiments with three dodder species, *Cuscuta campestris* YUNCKER, *C. epithymum* MURRAY, and *C. subinclusa* DUR. & HILG., had negative results. These experiments will not be described here.

Similarly preliminary experiments with the leafhoppers *Aphrodes bicinctus* SCHRK., *Euscelis* spp., and *Macropsis fuscula* ZETT. have not yet resulted in virus transmission.

In order to demonstrate clearly whether the disease is infectious or not, a number of graft experiments were carried out. However, grafting bilberry proved to be very difficult. In two experiments performed in the glasshouse February 20 and April 9, 1959, we succeeded in grafting diseased scions onto a number of healthy plants by means of the bottle-graft method. After some time some grafts died, but in February 1960 a number of them were still alive and developing as a broom on top of a healthy plant. In grafted plants the first symptoms of the disease appeared during January 1960, and in February the symptoms were conclusive (Fig. 2). February 22, 1960, the results were as follows: seven

out of eight plants grafted February 20, 1959, and six out of nine plants grafted April 9, 1959, showed a beginning distinct witches' broom growth. All 15 healthy, non-grafted control plants were entirely normal, whereas 13 diseased control plants did not show any sign of recovery.

These experiments clearly demonstrated the virus nature of the witches' broom disease of *Vaccinium myrtillus* in The Netherlands.

DISCUSSION

The identification of viruses that are not sap transmissible is generally difficult and often impossible. The viruses usually cannot be studied directly, but only indirectly on the basis of symptomatology, host range and vector-virus relationships. Therefore, the interrelation of witches' broom viruses described so far in the literature is an unsolved problem (cf. Bos, 1957). Even the possibility of proper cross protection tests seems to be lacking (VALENTA, 1959 a, b).

Until more research has been done on host range and vectors of the disease described in this paper, no accurate conclusion as to the exact identity of the virus is possible. The same holds for its relation to other witches' broom viruses occurring in The Netherlands, such as the witches' broom virus disease of raspberry ("Rubus stunt") (DE FLUITER & THUNG, 1951) and the witches' broom virus disease of clover (Bos, 1959). Since all the witches' broom viruses that have been studied in more detail appear not to be restricted to one plant species, a possible relationship between these diseases and the *Vaccinium* disease has not yet been excluded. Apparent restriction of one virus to certain crops may be due to an inability of the leafhopper vector to feed satisfactorily on different crops.

The disease may be related to „cranberry false blossom", a witches' broom virus disease of *Vaccinium macrocarpon* AIT, occurring in the U.S.A. This disease can be distinguished from that of *V. myrtillus*, however, by the presence of very striking floral abnormalities (antholysis). This difference might be due to the host plants. The American disease was described in detail for the first time by SHEAR in 1916. DOBROSKY (1929, 1931) first demonstrated its virus nature by transmitting the incitant by means of the leafhopper *Sclerorachus vaccinii* VAN DUZEE (= *Euscelis striatulus* FALL.). Later, by means of dodder, KUNKEL (1945) transmitted the virus to 28 plant species belonging to 10 different plant families.

The disease may also be related to the "blueberry stunt" virus disease of *Vaccinium australe* SMALL (= *V. corymbosum* L.) briefly described by WILCOX (1942). HUTCHINSON (1955) reported the leafhopper *Scaphytopius (Cloanthanus) magdalenensis* PROV. to be a vector of this virus¹.

In a paper read at the Plant Protection Congress in Berlin 1955, BLATTNÝ (1956) referred to a dangerous "little leaf disease" of *Vaccinium vitis-idaea* and especially of *V. myrtillus*, which he assumed to be of virus nature. The disease occurs in Bohemia, Czechoslovakia, where, especially in Southern Bohemia, it affects most of the bilberries.

¹ After the present paper had been accepted for publication, a more detailed description of this blueberry disease appeared in a publication of M. T. HUTCHINSON, A. C. GOHEEN & E. H. VARNEY on "Wild sources of blueberry stunt virus in New Jersey" (Phytopathology 50, 1960: 308-312). This description suggests blueberry stunt to be a different disease.

According to BLATTNY (personal communication, 1957), on the basis of symptoms in dried plant material and photographs which were sent to him by the present author, the Dutch disease was quite the same as that occurring in Czechoslovakia. Fresh material which was shown to the present author during a visit to Czechoslovakia in 1959 led to the same conclusion. The occurrence at two different sites in Europe suggests that the disease is more wide-spread than is known at present.

In this paper the virus nature of this European witches' broom disease of *Vaccinium* is demonstrated for the first time.

Several aspects of the disease are to be studied further.

SUMMARY

A description is given of a witches' broom disease of *Vaccinium myrtillus* L., occurring in the Netherlands. The symptoms appear to be identical to those of a "little leaf disease" of the same host and of *V. vitis-idaea* L. occurring prevalently in Bohemia (Czechoslovakia), according to BLATTNY (1956).

In this paper the virus nature of the disease was demonstrated for the first time.

The relation of this European virus disease to other witches' broom virus diseases and especially to the American "cranberry false blossom" and „blueberry stunt" is still uncertain. Both American viruses are leafhopper-borne, as are all witches' broom viruses.

SAMENVATTING

Een voor Nederland nieuwe heksenbezemziekte, namelijk in bosbes, *Vaccinium myrtillus* L., wordt kort beschreven. De ziekte komt vrij veel voor in de bossen bij Bennekom. Het ziektebeeld bestaat uit een opvallende heksenbezem-groei. De aangetaste planten kunnen, afhankelijk van hun ontwikkeling ten tijde van de besmetting, in uiteenlopende mate abnormaal sterk vertakt zijn, terwijl de nieuw gevormde takken korter blijven, bezet zijn met kleine bladeren en een steile, opgerichte stand hebben.

In de ernstigste vorm zijn de planten slechts ongeveer 5 cm hoog en hebben als geheel een zeer opvallende, bossige habitus (fig. 1). De bladeren zijn doorgaans lichter van kleur dan normaal en soms rood aangelopen. Aangetaste planten vormen in het geheel geen bloemen en daardoor ook geen vruchten meer.

De ziekteverschijnselen komen geheel overeen met een door BLATTNY (1956) in Tsjechoslowakije beschreven „kleinbladigheid" van de bosbes, welke soms ook bij *V. vitis-idaea* L. voorkomt en vooral in Bohemen zeer veel optreedt.

Door middel van enting werd de ziekte overgebracht op oorspronkelijk gezonde planten (fig. 2). Hiermee werd voor het eerst bewezen, dat deze in Europa voorkomende ziekte wordt veroorzaakt door een virus.

De verwantschap met andere heksenbezemvirusziekten en met de in Amerika optredende „cranberry false blossom disease" en de „blueberry stunt disease" is nog onzeker. De verwekkers van deze Amerikaanse ziekten zijn evenals alle heksenbezemvirussen niet over te brengen met sap of door contact en worden in de natuur door cicaden verspreid.



FIG. 1. Witches' broom diseased branches of bilberry (*Vaccinium myrtillus*). Left, two healthy branches. *Heksenbezemzieke takken van natuurlijk geïnfecteerde bosbesplanten* (*Vaccinium myrtillus*). Links, twee gezonde takken.



FIG. 2. Graft transmission of the witches' broom virus of bilberry (*Vaccinium myrtillus*). Left, healthy ungrafted plant; centre, plant grafted February 20, 1959, showing a beginning witches' broom growth; right, diseased plant. Photograph made February 2, 1960. In contrast to the diseased plant, the healthy plant lost its leaves during winter.

Overbrenging van het heksenbezemvirus van bosbes (*Vaccinium myrtillus*) door middel van enting. Links, gezonde, niet geënte plant; midden, plant geënt op 20 februari 1959, met beginnende heksenbezemgroei; rechts, zieke plant. Foto gemaakt op 2 februari 1960. In tegenstelling tot de zieke plant heeft de gezonde plant alle bladeren tijdens de winter verloren.

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